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Original article

Non-drug-related electrocardiographic features in animal models in safety pharmacology☆

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Abstract

No study of a test article is complete without attempting to determine its risk for production of toxicity to all important components of cardiovascular function (e.g., electrophysiological, mechanical, biochemical, baroreceptor). Electrocardiography is extremely useful for interrogating important electrophysiological properties: chronotropy (heart rate), dromotropy (conduction through the atria and ventricles, and through atrioventricular conduction), and predilection to produce arrhythmia, in particular, *torsade de pointes*. However, there are many factors that make electrocardiography less than optimal for detecting potential toxicological effects in studies of safety pharmacology. This paper will present examples of common difficulties in recording or in interpreting electrocardiograms, specifically due to artifacts in ECGs produced by the methods of electrocardiography, and by the "unusual" electrophysiology of the species/subject. One of the most contentious issues in electrocardiology is correction of QT for heart rate (Malik, M. (2001). Problems of heart rate correction in assessment of drug-induced QT interval prolongation. Journal of Cardiovascular Electrophysiology, 12, 411-420). This will not be discussed.

Keywords: Methods; Electrocardiography; ECG; Animal models

1. Methods of electrocardiography

The electrocardiograph (ECG) is a voltmeter designed to detect, amplify, and record voltages from the body surface; the voltages are produced by waves of depolarization and repolarization that traverse the heart (Einthoven, 1903; Lewis, 1912). Ideally, the ECG should register voltages from 0.05 to 5 mV and frequencies from 0.01 Hz (present in T-waves) to at least 60 Hz for dogs and monkeys and 150 Hz in smaller laboratory mammals (present in the QRS complex).

1.1. Frequency characteristics of the recorder

Fig. 1A and B illustrate a near "perfect" recording of the voltages generated by the heart, that is, all voltages of clinical importance.[1] The low-pass filter determines how high a frequency will be recorded faithfully by the ECG. Therefore, as the high-pass filter is decreased, the height of the R-wave decreases, because the R-wave contains relatively high-frequency information that requires a high-frequency response for faithful reproduction (Bailey et al., 1990; Dvir, Cilliers, & Lobetti, 2002; Jarrett & Flowers, 1991; Kulakowski, Counihan, Camm, & Mckenna, 1993; Rosas Peralta et al., 1996; Schrope, Fox, & Hahn, 1995).

☆ This article is dedicated to Dr. David K. Detweiler (Detweiler, 1981; Detweiler D.K., 1988; Detweiler et al., 1981; Detweiler D.R., 1988; Nahas et al., 2002), the father of modern comparative electrocardiology, who has contributed monumentally to our knowledge and the application of electrocardiology in safety pharmacology.

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[1] In fact extremely high-frequency (1 kHz), low-amplitude (<10 uV) voltages may occur at the end of the QRS complex and extend into the beginning of the ST-T. These "high-frequency components" indicate tardy and slow waves of depolarization "meandering" through ischemic ventricular myocardium, and the may indicate a predilection for the development of reentrant, potentially lethal ventricular arrhythmias (6,37,44).

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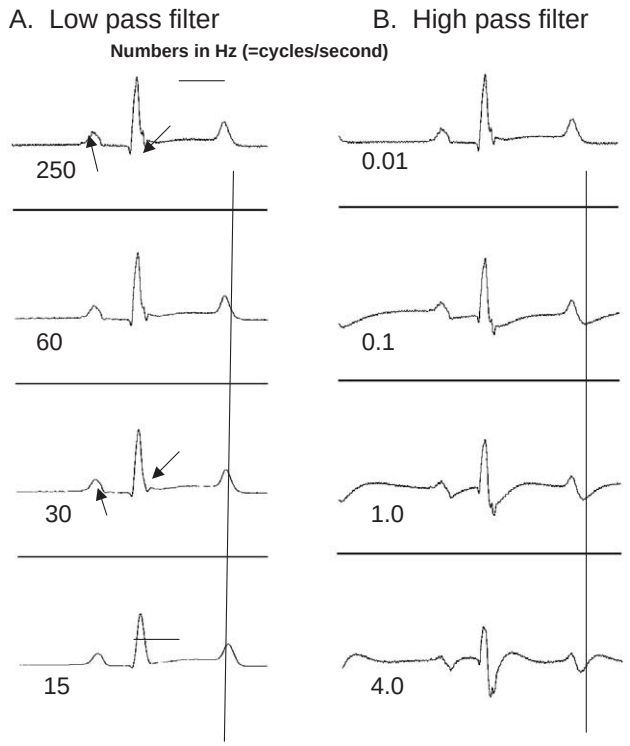

Fig. 1. Electrocardiogram (ECG) lead II displayed with various flow-pass (A) and high-pass (B) filtering. The two traces at the top are displayed with filtering between 0.01 and 250 Hz. On column A, the low-pass setting (representing the highest frequency component that is registered faithfully) is shown at the beginning of each ECG. Arrows point to high-frequency components to these ECGs, and it can be observed that these components disappear as the setting of the low-pass filter decreases from optimal (250 Hz). The horizontal line at the top of the 250 and 15 Hz recordings shows that the peak of the R-wave is blunted by the filtering. On column B, the high-pass setting (representing the lowest frequency component that may be recorded faithfully) is shown at the beginning of each ECG. It can be observed that less than optimal filtering (0.01 Hz) distorts the configurations of component deflections, in particular the end of the ST-T (shown by the vertical line).

Reduction of the low-pass filter decreases the relatively high-frequency components found in muscle tremor and 60 Hz artifact (which is good), but it also decreases the amplitude of high-frequency deflections (which is bad if the knowledge of the correct amplitude of a deflection is important).[2] This is reflected not only by reduction in the height of the R-wave, but also by the disappearance of the notches near the end of the R-wave and in the ascent of the P-wave.

The setting determines how low a frequency component may be recorded faithfully. This is demonstrated in Fig. 1B where the high-pass filter is increased from 0.01, to 0.1, then to 1.0, and finally to 4.0. If the top trace is what we desire to record, it can be observed that as the high-pass filter is increased, the ECG becomes distorted. Under these conditions, the P-wave becomes diphasic, the terminal, negative component to the QRS complex becomes very negative, and most importantly, the ST-T lengthens dramatically and the T-wave ends with a large negative component.

If one were to draw a vertical line from the ends of the ST-Ts obtained from a tracing displayed with optimal frequency characteristics (0.01 to 250 Hz), it would intersect with the ECGs taken with less-than-optimal frequency characteristics before the ST-T is completed. Thus, the QT measured from other than the ECG recorded with optimal characteristics will have an incorrect duration. This emphasizes the importance of recording with optimal filters, or at the very least, using ECGs with constant filtering from recording to recording.

An example of how less-than-optimal frequency characteristics of the recorder alter the configuration of the ECG is shown in Fig. 2. In this case, some T-waves will appear to end much later than others because of a negative deflection of long duration following the end of the true T-wave. This is an artifact due to an increase in the high-pass filter. Whereas the filter minimizes a "wandering" baseline, it does distort the true voltages.

The ideal ECG will pass frequencies between 0.01 and 60 Hz, but the smaller the animal, the higher the frequency components to the ECG Thus, the ECG of a mouse or rat must be recorded with an ECG possessing a low-pass filter of well over 120 Hz.[3]

Many ECG artifacts arise from faulty coupling of the electrodes (Almasi, Schmitt, & Jankus, 1970) used to couple the skin to the electrocardiograph. The better the coupling, i.e., the lower the resistance, the less likely the 60 Hz artifact will be observed.[4] Artifacts resulting from electrodes may be reduced by using electrode plates or pads that contact the skin over a relatively large area.[5]

2. Common ECG artifacts

2.1. Error in electrode placement

A very common ECG artifact occurs when the technician applies an electrode, intended for the right thoracic limb, to the left thoracic limb. This almost always can be identified (Fig. 3) by observing a negative P-wave in lead I. The P-wave in lead I usually is positive with a sinus rhythm. In this circumstance, the trace may be interpreted without error by merely reading it upside-down.[6]

[2] It is exceedingly rare that knowledge of amplitudes of electrocardiographic deflections are useful in non-clinical studies in safety pharmacology.

[3] Of little interest in safety pharmacology but of great interest in equine medicine, the ECG from a horse with a very slow heart rate and ECG deflections of long duration, it might be necessary to lower the high-pass filter to 0.005 Hz in order to obtain a faithful registration of voltages (in particular the ST-T) produced by the heart.

[4] Alcohol is an insulator; ketchup is much better (it's very salty); commercially available electrode paste is best.

[5] Alligator clips may be fast to apply, but they are often corroded, injure the skin, are painful, and worst of all make contact with only a small area.

[6] Of course amplitudes are rarely–if ever–of value in establishing a toxic effect in studies in safety pharmacology.